CLAIMS

What Is Claimed:

1. A method of identifying agents that modulate the cleavage of APP by a β-secretase comprising:

(a) providing a chimeric molecule, wherein the chimeric molecule includes a transmembrane region, an APP β-secretase cleavage site, and an extracellular region;

(b) contacting the chimeric molecule with a β-secretase in the presence and absence of at least one potential cleavage modulating agent; and

(c) identifying occurrences of cleavage of the chimeric molecule;

wherein a difference in cleavage in the presence of the agent relative to cleavage in the absence of the agent is indicative of a cleavage modulating agent.

2. The method of claim 1, wherein the β-secretase cleavage site is the amino acid sequence EVKMDAE.

3. The method of claim 1, wherein the β-secretase cleavage site is the amino acid sequence EVNLDAE.

4. The method of claim 1, wherein the chimeric protein is expressed from an expression vector.

5. The method of claim 4, wherein the chimeric protein is expressed in a host cell.

6. The method of claim 5, wherein the host cell expresses an active β-secretase enzyme.

7. The method of claim 6, wherein the host cell expresses an endogenous β-secretase enzyme.

8. The method of claim 6, wherein the host cell comprises an expression vector that expresses β-secretase.

9. The method of claim 1, wherein identifying occurrences of cleavage comprises detection of a cleaved extracellular region.

10. The method of claim 1, wherein the extracellular region includes the central APP domain (CAD).

11. The method of claim 1, wherein the extracellular region binds F-spondin.

12. A method of identifying agents that modulate the cleavage of APP by a β-secretase comprising:

(a) providing a chimeric molecule, wherein the chimeric molecule includes a transmembrane region with a γ-secretase cleavage site, a β-secretase cleavage site, and an APP C-terminal cytoplasmic tail modified to allow detection of nuclear localization;

(b) contacting the chimeric molecule with a β-secretase in the presence and absence of at least one potential modulating agent;

(c) contacting the chimeric molecule with a γ-secretase; and

(d) identifying occurrences of cleavage by measuring nuclear localization of the C-terminal cytoplasmic tail

wherein a difference in cleavage in the presence of the agent relative to cleavage in the absence of the agent is indicative of a cleavage modulating agent.

13. The method of claim 12, wherein the β-secretase cleavage site is the amino acid sequence EVKMDAE.

14. The method of claim 12, wherein the chimeric protein is expressed from an expression vector.

15. The method of claim 14, wherein the protein is expressed in a host cell.

16. The method of claim 15, wherein the host cell expresses an active β-secretase enzyme.

17. The method of claim 15, wherein the host cell expresses an endogenous β-secretase enzyme.

18. The method of claim 15, wherein the host cell comprises an expression vector that expresses β-secretase.

19. A method of identifying agents that specifically modulate the cleavage of APP by a β-secretase with respect to cleavage of APLP comprising contacting an APLP with β-secretase in the presence and absence of a modulator of β-secretase cleavage of APP, wherein lack of a significant difference in cleavage of the APLP in the presence and absence of the modulator is indicative of a specific modulator of β-cleavage of APP.

20. The method of claim 19, wherein the APLP is APLP1.

21. The method of claim 19, wherein the APLP is APLP2.

22. A composition comprising a polypeptide substrate for cleavage by β-secretase comprising a transmembrane region and an exogenous APP β-secretase cleavage site inserted into the polypeptide near the transmembrane region.

23. The composition of claim 22, wherein the exogenous β-secreatase cleavage site is inserted from 1 to 100 residues from the transmembrane region.

24. The composition of claim 22, wherein the exogenous β-secreatase cleavage site is inserted from 10 to 90 residues from the transmembrane region.

25. The composition of claim 22, wherein the exogenous β-secreatase cleavage site is inserted from 40 to 50 residues from the transmembrane region.

26. The composition of claim 22, wherein the β-secreatase cleavage site is the amino acid sequence EVKMDAE.

27. An isolated nucleic acid encoding the polypeptide substrate of claim 22.

28. A host cell comprising the nucleic acid of claim 27.

29. The host cell of claim 28, further defined as a mammalian cell.

30. A composition comprising a polypeptide substrate for cleavage by β-secretase comprising a transmembrane region and an exogenous APLP1 β-secretase cleavage site inserted into the polypeptide near the transmembrane region.

31. The composition of claim 30, wherein the exogenous β-secreatase cleavage site is inserted from 1 to 100 residues from the transmembrane region.

32. The composition of claim 30, wherein the exogenous β-secreatase cleavage site is inserted from 10 to 90 residues from the transmembrane region.

33. The composition of claim 30, wherein the exogenous β-secreatase cleavage site is inserted from 40 to 50 residues from the transmembrane region.

34. The composition of claim 30, wherein the β-secreatase cleavage site is the amino acid sequence DELAPAGTGVSRE.

35. An isolated nucleic acid encoding the polypeptide substrate of claim 30.

36. A host cell comprising the nucleic acid of claim 35.

37. The host cell of claim 36, further defined as a mammalian cell.

38. A method of identifying agents that modulate the cleavage of APP like proteins by a β-secretase comprising:

(a) providing a chimeric molecule, wherein the chimeric molecule includes a transmembrane region, an APLP β-secretase cleavage site, and an extracellular region;

(b) contacting the chimeric molecule with a β-secretase in the presence and absence of at least one potential cleavage modulating agent; and

(c) identifying occurrences of cleavage of the chimeric molecule;

wherein a difference in cleavage in the presence of the agent relative to cleavage in the absence of the agent is indicative of a cleavage modulating agent.

39. The method of claim 38, wherein the APLP β-secretase cleavage site is an APLP1 cleavage site.

40. The method of claim 38, wherein the APLP β-secretase cleavage site is an APLP2 cleavage site.

41. The method of claim 38, wherein the chimeric protein is expressed from an expression vector.

42. The method of claim 41, wherein the protein is expressed in a host cell.

43. The method of claim 42, wherein the host cell expresses an active β-secretase enzyme.

44. The method of claim 43, wherein the host cell expresses an endogenous β-secretase enzyme.

45. The method of claim43, wherein the host cell comprises an expression vector that expresses β-secretase.

46. The method of claim 38, wherein identifying occurrences of cleavage comprises detection of a cleaved extracellular region.

47. The method of claim 38, wherein the extracellular region includes the central APP domain (CAD).

48. The method of claim 38, wherein the extracellular region binds F-spondin.